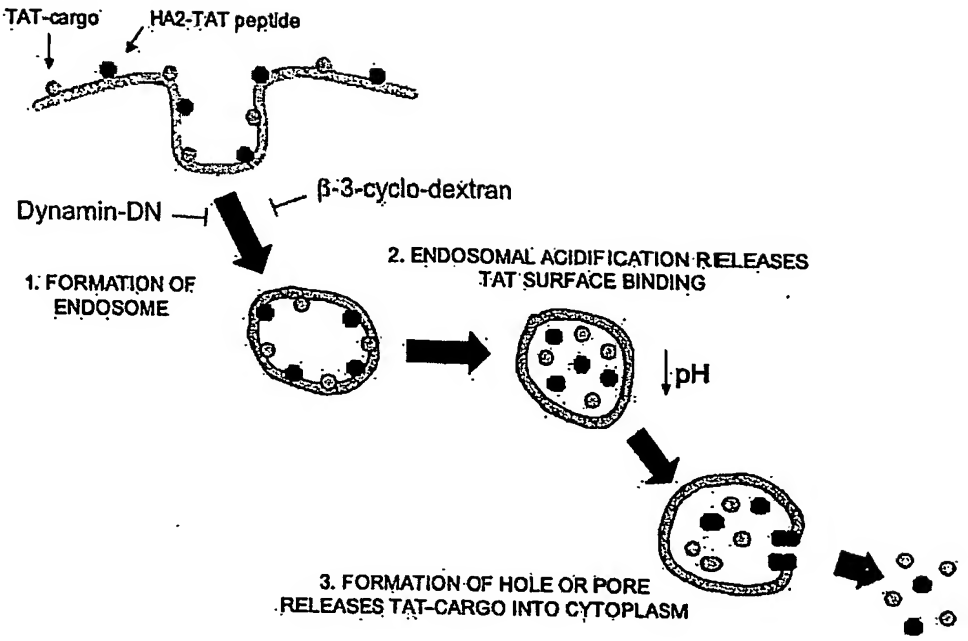

TAT-cargo
HA2-TAT peptide
Dynamin-DN
β-3-cyclo-dextran
1. FORMATION OF ENDOSOME
2. ENDOSOMAL ACIDIFICATION RELEASES TAT SURFACE BINDING
pH
3. FORMATION OF HOLE OR PORE RELEASES TAT-CARGO INTO CYTOPLASM

FIG. 1

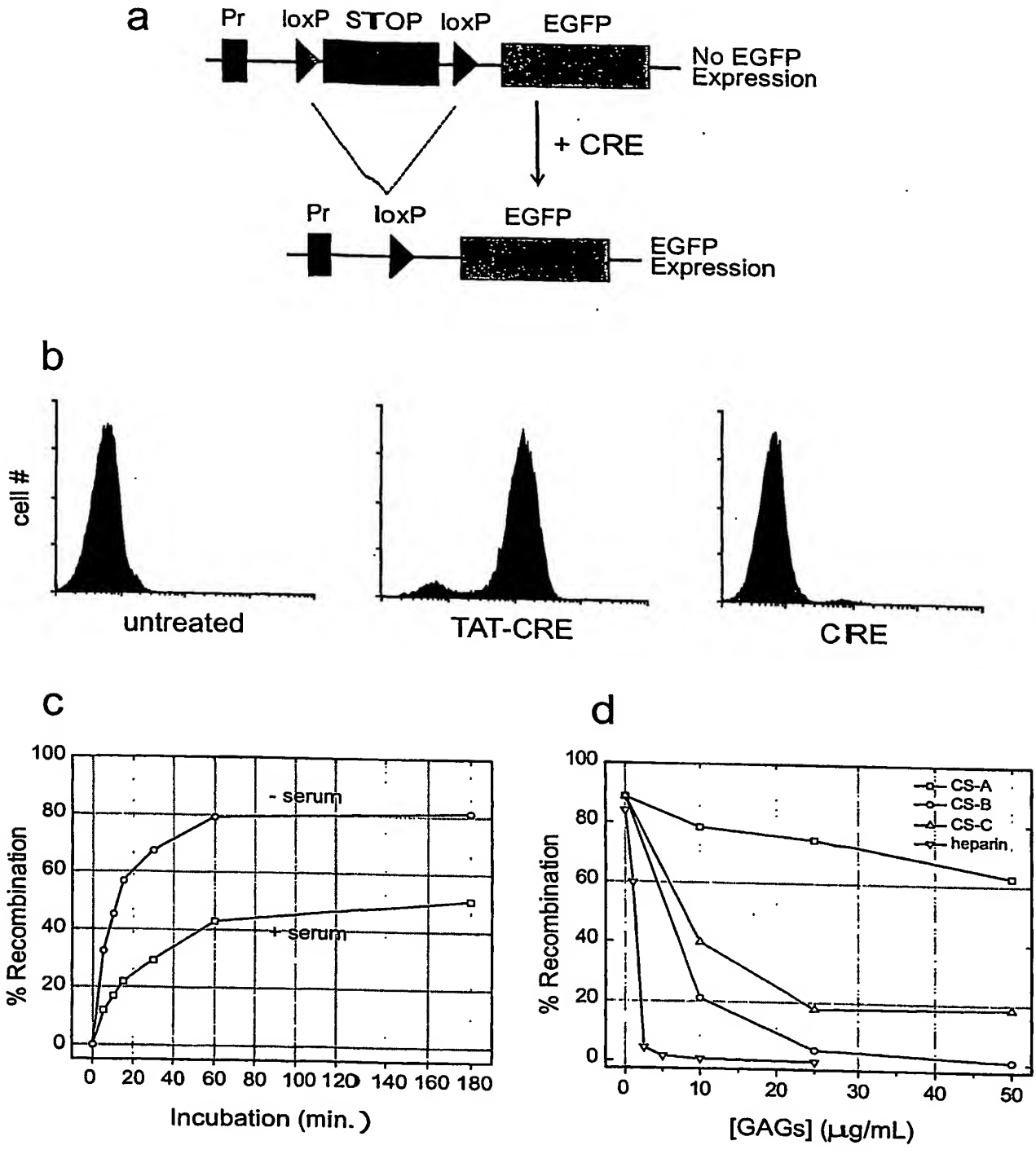

a
Pr
loxP
STOP
loxP
EGFP
No EGFP Expression
+ CRE
Pr
loxP
EGFP
EGFP Expression
b
cell #
untreated
TAT-CRE
CRE
c
% Recombination
- serum
+ serum
Incubation (min.)
d
% Recombination
CS-A
CS-B
CS-C
heparin
[GAGs] (µg/mL)

FIG. 2A-D

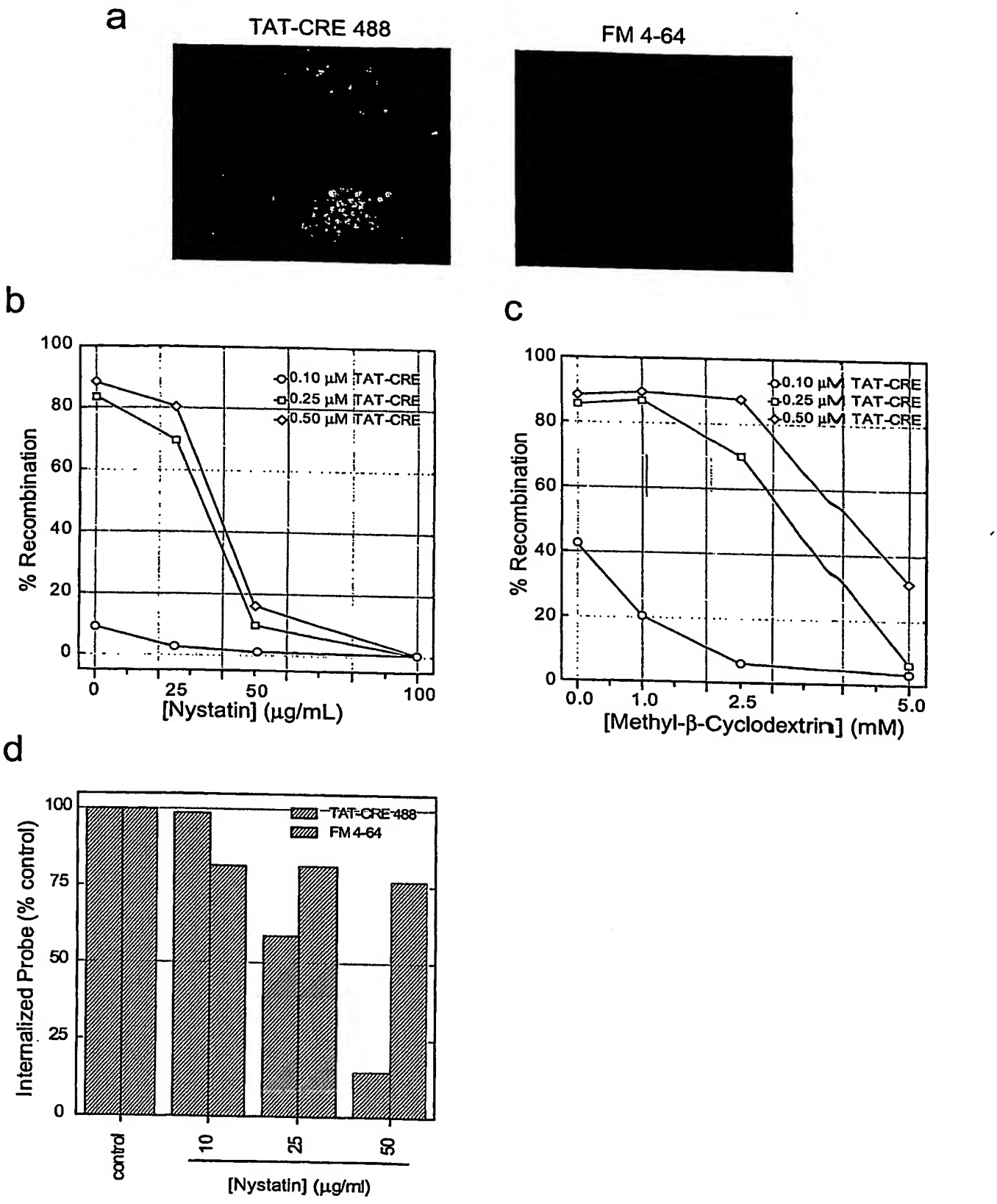


FIG. 3A-D

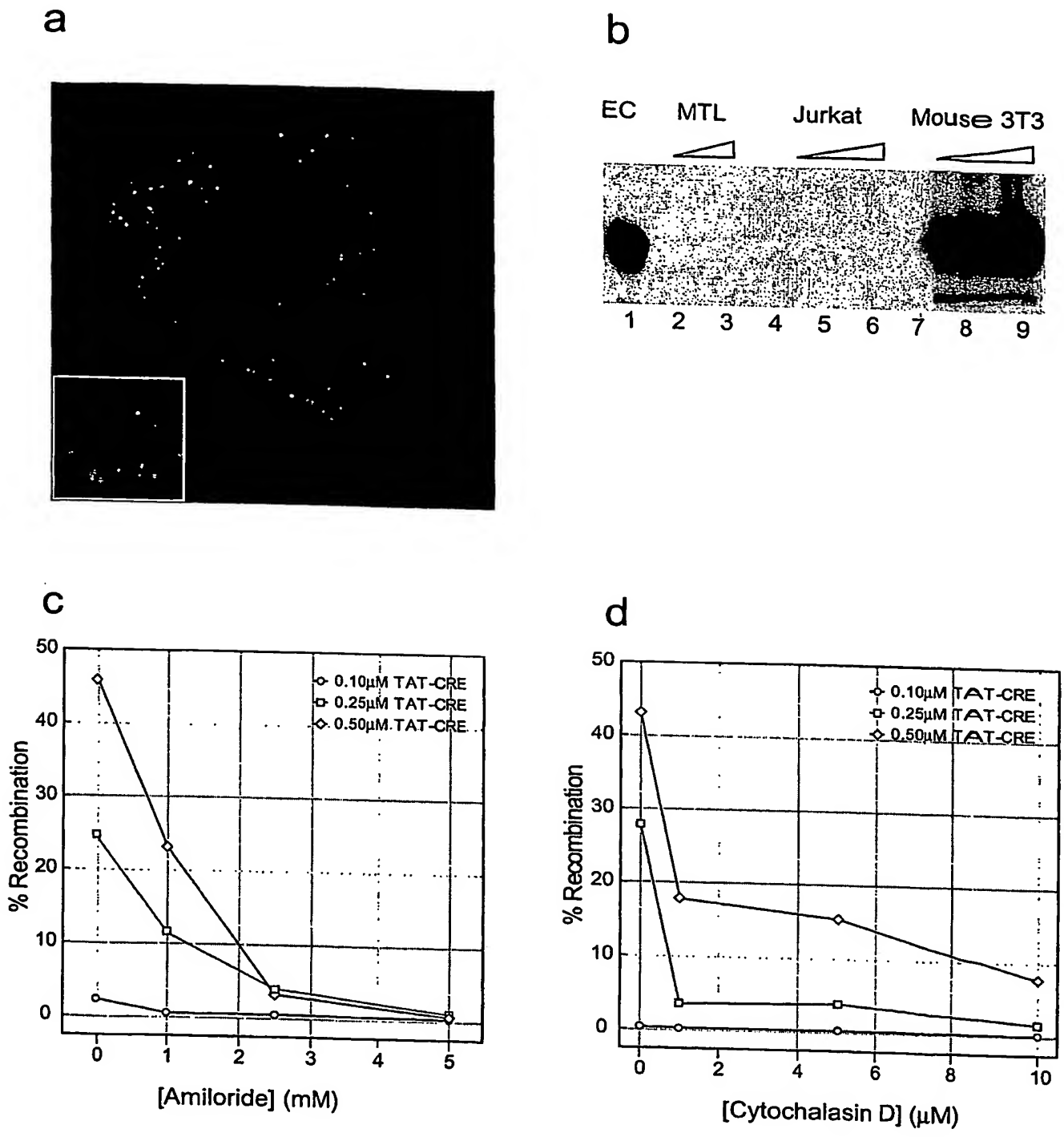


FIG. 4A-D

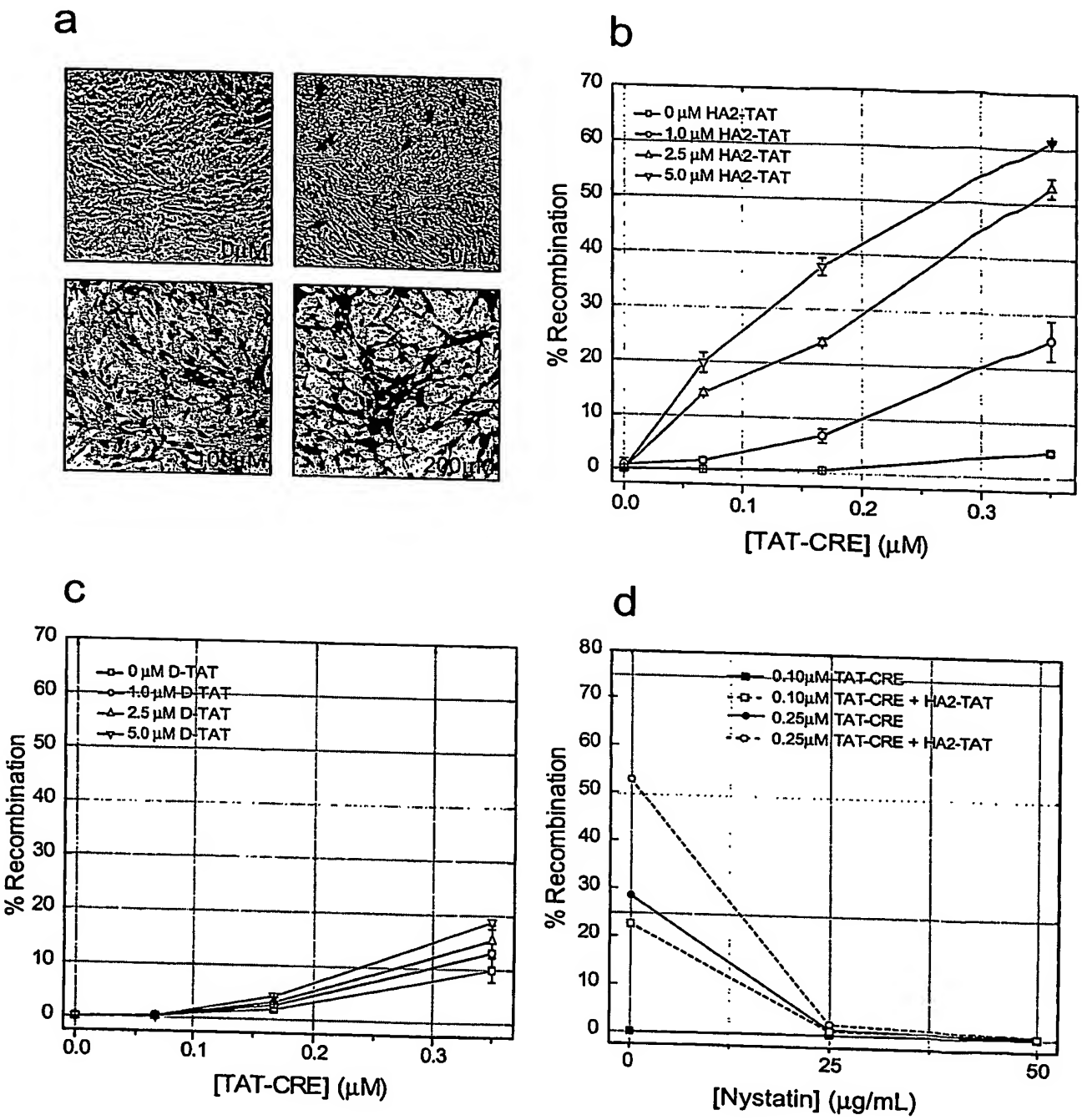

a
0μM
50μM
100μM
200μM
b
0 μM HA2-TAT
1.0 μM HA2-TAT
2.5 μM HA2-TAT
5.0 μM HA2-TAT
% Recombination
[TAT-CRE] (μM)
c
0 μM D-TAT
1.0 μM D-TAT
2.5 μM D-TAT
5.0 μM D-TAT
% Recombination
[TAT-CRE] (μM)
d
0.10μM TAT-CRE
0.10μM TAT-CRE + HA2-TAT
0.25μM TAT-CRE
0.25μM TAT-CRE + HA2-TAT
% Recombination
[Nystatin] (μg/mL)

FIG. 5A-D

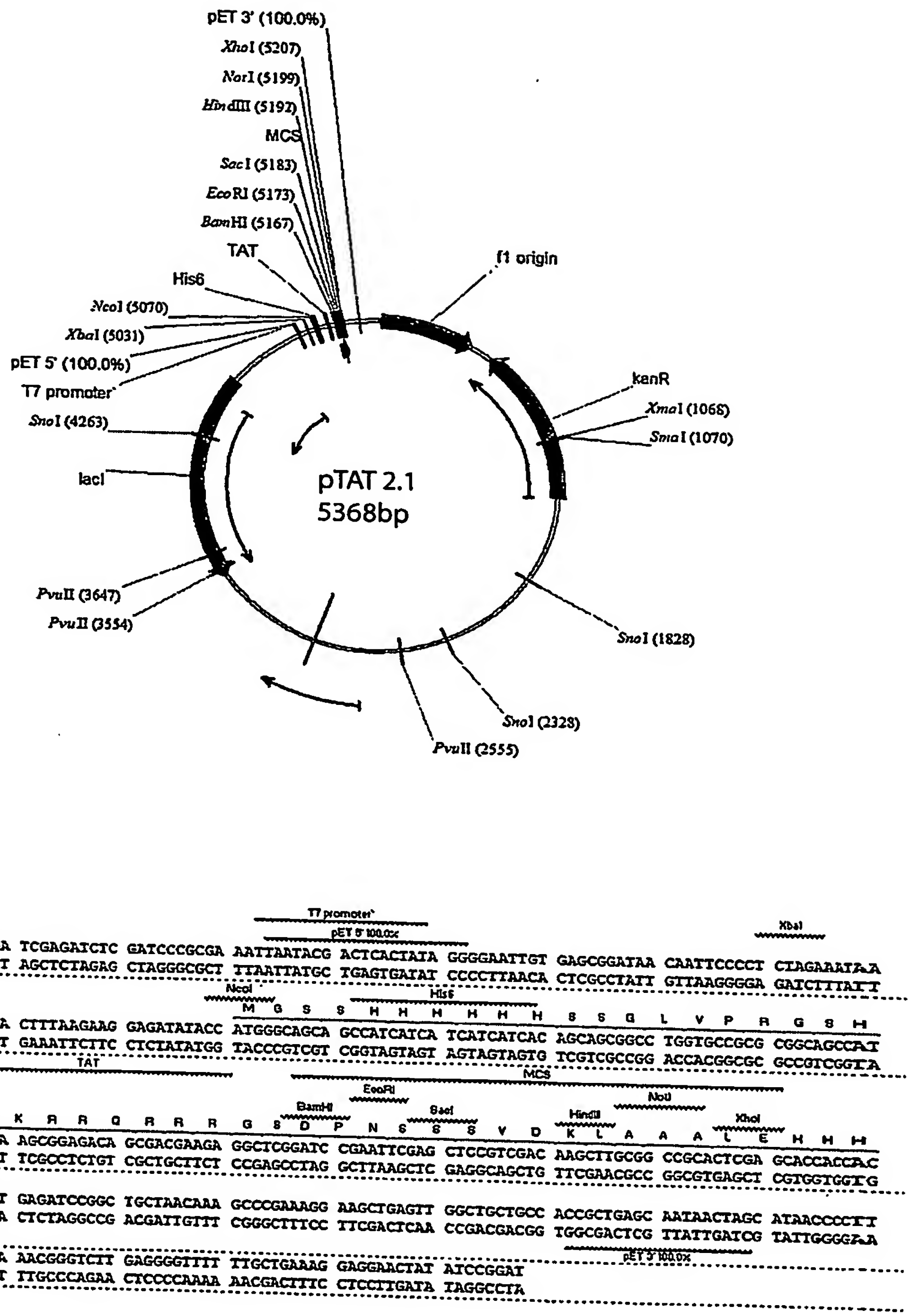
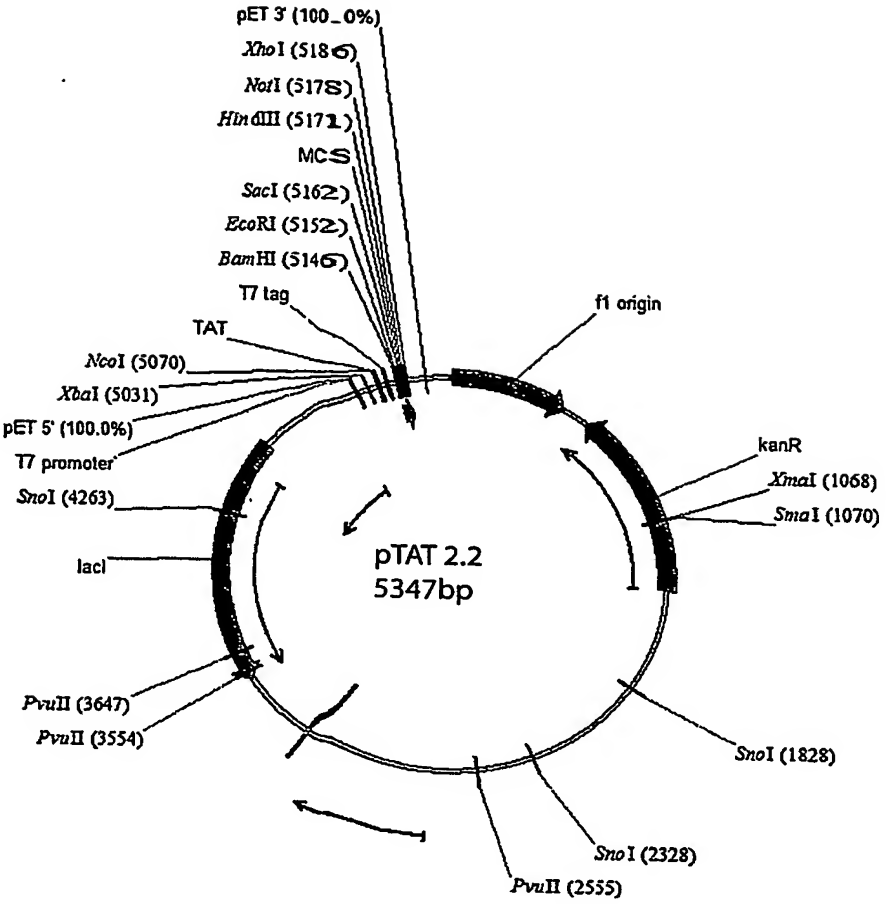

pET 3' (100.0%)
XhoI (5207)
NotI (5199)
HindIII (5192)
MCS
SacI (5183)
EcoRI (5173)
BamHI (5167)
TAT
His6
NcoI (5070)
XbaI (5031)
pET 5' (100.0%)
T7 promoter
SnoI (4263)
lacI
PvuII (3647)
PvuII (3554)
f1 origin
kanR
XmaI (1068)
SmaI (1070)
SnoI (1828)
SnoI (2328)
PvuII (2555)
pTAT 2.1
5368bp
T7 promoter
pET 5' 100.0%
XbaI
4951 GCGTAGAGGA TCGAGATCTC GATCCCGCGA AATTAATACG ACTCACTATA GGGGAATTGT GAGCGGATAA CAATTCCCCT CTAGAAATAA
CGCATCTCCT AGCTCTAGAG CTAGGGCGCT TTAATTATGC TGAGTGATAT CCCCTTAACA CTCGCCTATT GTTAAGGGGA GATCTTTATT
NcoI
His6
M G S S H H H H H H S S G L V P R G S H
5041 TTTTGTTTAA CTTTAAGAAG GAGATATACC ATGGGCAGCA GCCATCATCA TCATCATCAC AGCAGCGGCC TGGTGCCGCG CGGCAGCCAT
AAAACAAATT GAAATTCTTC CTCTATATGG TACCCGTCGT CGGTAGTAGT AGTAGTAGTG TCGTCGCCGG ACCACGGCGC GCCGTCGGTA
TAT
MCS
EcoRI
NotI
BamHI
SacI
HindIII
XhoI
M R K K R R Q R R R G S D P N S S S V D K L A A A L E H H H
5131 ATGAGGAAGA AGCGGAGACA GCGACGAAGA GGCTCGGATC CGAATTCGAG CTCCGTCGAC AAGCTTGCGG CCGCACTCGA GCACCACCAC
TACTCCTTCT TCGCCTCTGT CGCTGCTTCT CCGAGCCTAG GCTTAAGCTC GAGGCAGCTG TTCGAACGCC GGCGTGAGCT CGTGGTGGTG
H H H
5221 CACCACCACT GAGATCCGGC TGCTAACAAA GCCCGAAAGG AAGCTGAGTT GGCTGCTGCC ACCGCTGAGC AATAACTAGC ATAACCCCTT
GTGGTGGTGA CTCTAGGCCG ACGATTGTTT CGGGCTTTCC TTCGACTCAA CCGACGACGG TGGCGACTCG TTATTGATCG TATTGGGGAA
pET 3' 100.0%
5311 GGGGCCTCTA AACGGGTCTT GAGGGGTTTT TTGCTGAAAG GAGGAACTAT ATCCGGAT
CCCCGGAGAT TTGCCCAGAA CTCCCCAAAA AACGACTTTC CTCCTTGATA TAGGCCTA

FIG. 6


pET 3' (100_0%)
XhoI (5186)
NotI (5178)
HindIII (5171)
MCS
SacI (5162)
EcoRI (5152)
BamHI (5146)
T7 tag
TAT
NcoI (5070)
XbaI (5031)
pET 5' (100.0%)
T7 promoter
SnoI (4263)
lacI
PvuII (3647)
PvuII (3554)
f1 origin
kanR
XmaI (1068)
SmaI (1070)
pTAT 2.2
5347bp
SnoI (1828)
SnoI (2328)
PvuII (2555)

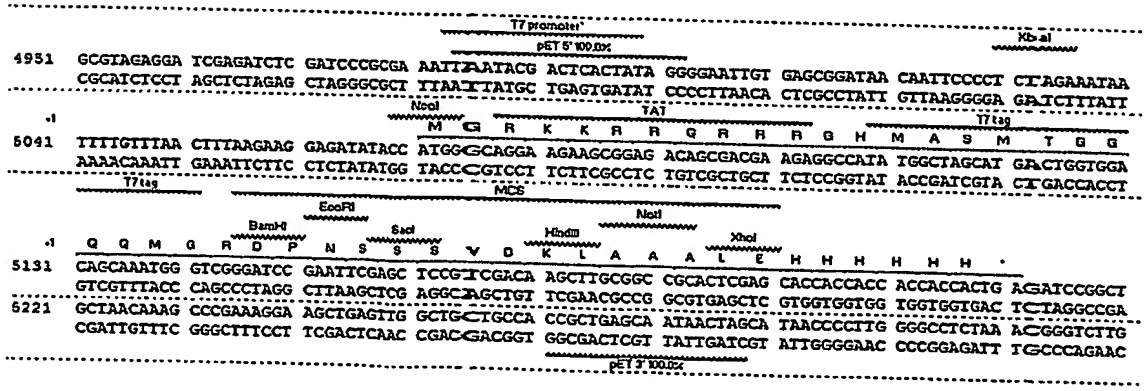
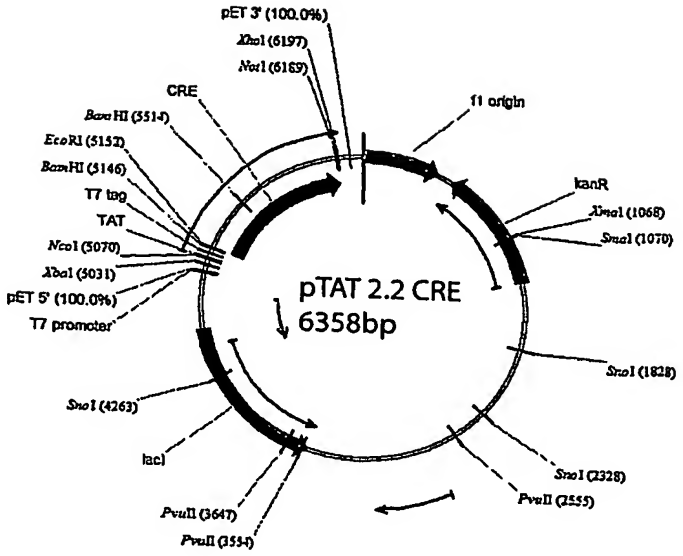

T7 promoter
pET 5' 100.0%
XbaI
4951 GCGTAGAGGA TCGAGATCTC GATCCCGCGA AATTAATACG ACTCACTATA GGGGAATTGT GAGCGGATAA CAATTCCCCT CTAGAAATAA
CGCATCTCCT AGCTCTAGAG CTAGGGCGCT TTAATTATGC TGAGTGATAT CCCCTTAACA CTCGCCTATT GTTAAGGGGA GATCTTTATT
NcoI
TAT
T7 tag
-1 M G R K K R R Q R R R G H M A S M T G G
5041 TTTTGTTTAA CTTTAAGAAG GAGATATACC ATGGGCAGGA AGAAGCGGAG ACAGCGACGA AGAGGCCATA TGGCTAGCAT GACTGGTGGA
AAAACAAATT GAAATTCTTC CTCTATATGG TACCCGTCCT TCTTCGCCTC TGTCGCTGCT TCTCCGGTAT ACCGATCGTA CTGACCACCT
T7 tag
MCS
EcoRI
BamHI
SacI
HindIII
NotI
XhoI
-1 Q Q M G R D P N S S S V D K L A A A L E H H H H H H *
5131 CAGCAAATGG GTCGGGATCC GAATTCGAGC TCCGTCGACA AGCTTGCGGC CGCACTCGAG CACCACCACC ACCACCACTG AGATCCGGCT
GTCGTTTACC CAGCCCTAGG CTTAAGCTCG AGGCAGCTGT TCGAACGCCG GCGTGAGCTC GTGGTGGTGG TGGTGGTGAC TCTAGGCCGA
5221 GCTAACAAAG CCCGAAAGGA AGCTGAGTTG GCTGCTGCCA CCGCTGAGCA ATAACTAGCA TAACCCCTTG GGGCCTCTAA ACGGGTCTTG
CGATTGTTTC GGGCTTTCCT TCGACTCAAC CGACGACGGT GGCGACTCGT TATTGATCGT ATTGGGGAAC CCCGGAGATT TGCCCAGAAC
pET 3' 100.0%

FIG. 7


pET 3' (100.0%)
XhoI (6197)
NotI (6169)
CRE
f1 origin
BamHI (5514)
EcoRI (5152)
BamHI (5146)
T7 tag
TAT
NcoI (5070)
XbaI (5031)
pET 5' (100.0%)
T7 promoter
pTAT 2.2 CRE
6358bp
kanR
XmaI (1068)
SmaI (1070)
SnoI (1828)
SnoI (2328)
PvuII (2555)
SnoI (4263)
lacI
PvuII (3647)
PvuII (3554)

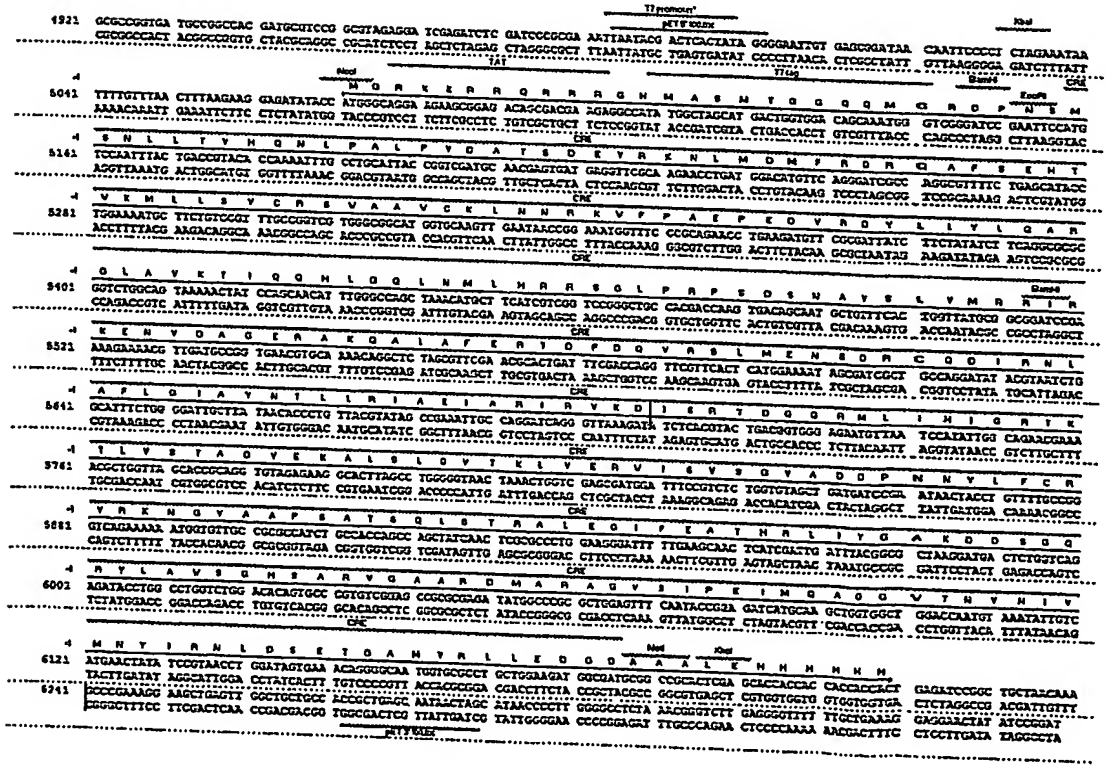


FIG. 8